Fatty acids profile and alteration of lemon seeds extract (*Citrus limon*) added to soybean oil under thermoxidation

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Abstract This paper aimed at evaluating fatty acids profile and the total alteration of lemon seeds extract added to soybean oil under thermoxidation, verifying the isolated and synergistic effect of these antioxidants. Therefore, Control treatments, LSE (2,400 mg/kg Lemon Seeds Extract), TBHQ (mg/kg), Mixture 1 (LSE+50 mg/kg TBHQ) and Mixture 2 (LSE+25 mg/kg TBHQ) were subjected to 180°C for 20 h. Samples were taken at 0, 5, 10, 15 and 20 h intervals and analyzed as for fatty acid profile and total polar compounds. Results were subjected to variance analyses and Tukey tests at a 5% significance level. An increase in the percentage of saturated fatty acids and mono-unsaturated, and decrease in polyunsaturated fatty acids was observed, regardless of the treatments studied. For total polar compounds, it was verified that Mixtures 1 and 2 presented values lower than 25% with 20 h of heating, not surpassing the limits established in many countries for disposal of oils and fats under high temperatures, thus proving the synergistic effect of antioxidants.

Keywords Fatty acids · Total polar compounds · Gas chromatography · Thermoxidation

Introduction

Oils and fats are predominantly triesters of glycerol and fatty acids, called triacylglycerols. In general, saturated fatty acids tend to raise blood cholesterol in all fractions of lipoproteins. Moreover, the consumption of polyunsaturated fatty acids

food sources, especially omega-3 and omega-6 is associated with a reduced risk of developing various diseases such as atherosclerosis and cardiovascular diseases.

The composition of fatty acids in food, principally grain and flax seed is of great importance, especially the families of polyunsaturated, omega-3 and omega-6, which are attributed to be beneficial for the human body (Rajiv et al. 2011). Omega-3 essential fatty acid α -linolenic (C18: 3, ω -3) family, by elongation and desaturation, generates eicosapentaenoic acid (EPA - C20: 5) and docosahexaenoic (DHA - C22: 6). Omega-6 essential fatty acid linoleic acid family, can lead to arachidonic acid (Rubio-Rodríguez et al. 2010).

The thermoxidation process is used extensively to simulate the frying process. It consists of subjecting oils and fats to high temperatures without the presence of food. In the absence of moisture and other food borne compounds, temperature and oxygen from the air are the main variables to be considered (Shyamala et al. 2005).

Frying is a complex process in which food is submerged into hot oil that, acting as a mean of heat transfer, provides the product with features such as nice color, flavor, texture and palatability. Besides these positive changes, the process is also responsible for the occurrence of degradation reactions, which modify the functional and nutritional qualities of food, reaching levels at which the product becomes unfit for consumption and without the desired quality (Zaiifar et al. 2008).

By heating oil, a series of reactions produces many degradation compounds, and more than 400 different chemical compounds have been identified in damaged heated oil (Almeida-Doria and Regitano-D'arce 2000). Peroxides, free fatty acids, oxidized fatty acids, polymeric compounds, polar compounds (alcohols, aldehydes, ketones, partial glycerides, dimers) are the degradation products formed in the frying oil (Khan et al. 2011).

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Non-volatile degradation products, which remain in the oil, promote its further deterioration and are responsible for changes in the physical and chemical properties of this oil. The most frequently physical changes observed are increase in viscosity, change of color and foaming. As a result of chemical changes, there are free fatty acids increase, carbonyl compounds, products of high molecular weight, reduction of unsaturations, among other effects (Reische et al. 2002).

The degree of oil unsaturation has long been regarded as one of the most important factors, due to different reactivity of unsaturated fatty acids. According to Paul and Mittal (1997), vegetable oils have high levels of monounsaturated and polyunsaturated fatty acids. Thus, they are more susceptible to oxidative changes than oils with higher amounts of saturated fatty acids (Lolos et al. 1999). Soon they are rancid at room temperature and with lower quality for operations of heating with short periods of use. Also, they become unsuitable for use in food products that require longer shelf life.

There is a wide variety of analytical methods used for evaluation of oils and fats under high temperatures (Stevenson et al. 1984; White 1991; Bastida and Sánchez-Muniz 2001). However, the determination of total polar compounds by column chromatography has been reported by several authors as one of the best methods for determining the state of alteration of heating oil (Pérez-Camino et al. 1988; Marmesat et al. 2007).

Determination of the total amount of changed products originated as a consequence of the heating, is the basis of oil use limitations in many countries, established around 25% of polar compounds (Stevenson et al. 1984) because the higher the polar fraction, the worse the oil quality.

It is understood by total polar compounds all those with polarity greater than triacylglycerols corresponding to non-volatile, and resulting from oxidative, thermal and hydrolytic modifications (Dobarganes and Márquez-Ruiz 1998).

This way, the objectives of this work were to evaluate the fatty acids profile and the total alteration of soybean oil thermoxidation added to lemon seeds extract, verifying the isolated and synergistic effect of these natural and synthetic antioxidants.

Experimental procedures

Materials

Lemon seeds

In order to perform this study, we bought mature *galego* lemon from a crop in the Southeastern region of São Paulo, in the city of Itajobi, Brazil, harvested in January 2007. The lemons were cut in half and the seeds were manually

removed and, slightly washed in distilled water in order to remove residues of pulp and soluble sugars from the fruit. Seeds were dried in a kiln, with forced air circulation, at 45°C for 24 h to reduce the humidity content to less than 10%. After this, they were stored in plastic containers, closed with screw caps duly labeled for later analysis.

Oil

In order to carry out the experimental trial, refined soybean oil without the addition of synthetic antioxidants (TBHQ and citric acid) was used. Bottles containing 900 mL of soybean oil processed by the company Cargill Agrícola S/A, Uberlândia-Brasil were used.

Antioxidants

The synthetic antioxidant used was tert-butylhydroquinone (TBHQ), powdered, supplied by Danisco S/A.

In order to obtain the methanolic extract, dehydrated and crushed lemon seeds (10 g) were kept in constant agitation with methanol (100 mL) at 25±2°C, for 6 h and, afterwards, the mixture was centrifuged at 3,000 rpm, for 10 min. After the supernatant were transferred, the precipitate was again subjected to the process of extraction in the same conditions previously described, and the supernatant resulting from the extraction were combined. After that, the solvent used to obtain methanolic extract was removed over reduced pressure at 40°C. The dried extract was weighed and resuspended in methanol, obtaining a stock solution containing 1 g of methanolic extract for each 10 g of methanol solvent (1:10), used for direct application in the soybean oil.

Experimental test

The following treatment underwent thermoxidation, conducted in two repetitions: i) soybean oil without the addition of synthetic antioxidants and citric acid (Control), ii) soybean oil with the addition of 2,400 mg/kg of lemon seeds extract (LSE), iii) soybean oil with the addition of 50 mg/kg of TBHQ (TBHQ), iv) soybean oil with the addition of antioxidants mixture, in other words, 2,400 mg/kg of lemon seeds extract and 50 mg/kg of TBHQ (Mixture 1), and v) soybean oil with the addition of antioxidants mixture, in other words 2,400 mg/kg of lemon seeds extract and 25 mg/kg of TBHQ (Mixture 2). The value of 50 mg/kg of synthetic antioxidant TBHQ was defined, since it is the most common concentration of this antioxidant added to soybean oil by the industries.

The experimental trial was done in heated sheet, using beakers of 50 mL containing a 30 mL sample with surface/

Table 1 Fatty acid composition of control during thermoxidation at 180°C

	Fatty acids (%)	Heating times (h)*				
		0	5	10	15	20
	C14:0	0.05±0.02	0.27±0.02	0.29±0.03	0.42±0.02	0.46±0.04
	C16:0	10.5±0.01	10.9±0.03	12.0±0.04	13.1±0.03	13.3±0.03
* Mean ± standard deviation (n=2)	C16:1	0.13±0.03	0.24±0.03	0.25±0.04	0.32±0.04	0.33±0.02
a, b... (line)-averages followed by the same small letter do not differ by the Tukey test ($P>0.05$)	C18:0	1.2±0.03	2.2±0.02	2.4±0.04	3.3±0.05	3.4±0.02
	C18:1	18.9±0.02	19.9±0.04	21.1±0.05	22.9±0.05	24.5±0.01
	C18:2	58.7±0.04	57.8±0.04	56.1±0.01	54.4±0.03	52.9±0.03
C14:0, myristic acid; C16:0, palmitic acid; C16:1, palmitoleic acid; C18:0, stearic acid; C18:1, oleic acid; C18:2, linoleic acid; C18:3, α -linolenic acid; C20:0, arachidic acid; C20:1, eicosenoic acid; C22:0, behenic acid; AGS, saturated fatty acids; AGM, mono-unsaturated fatty acids; AGP, polyunsaturated fatty acids; NI, no identification	C18:3	7.8±0.03	5.9±0.04	4.7±0.01	2.8±0.03	2.0±0.05
	C20:0	–	0.14±0.03	0.18±0.03	0.26±0.01	0.31±0.07
	C20:1	–	0.03±0.02	0.03±0.02	0.08±0.03	0.09±0.07
	C22:0	–	0.15±0.01	0.22±0.03	0.35±0.02	0.39±0.04
	AGS	11.7 ^c	13.6 ^d	15.1 ^c	17.4 ^b	17.8 ^a
	AGM	19.0 ^c	20.2 ^d	21.3 ^c	23.3 ^b	25.0 ^a
	AGP	66.5 ^a	63.6 ^b	60.7 ^c	57.1 ^d	54.9 ^c
	NI	2.8	2.6	2.9	2.2	2.4

volume relation of 0.4/cm. The temperature was monitored at 180±5°C.

The trial was conducted in a discontinuous way, 10 h of heating were performed per day, and the samples were taken in 0, 5, 10, 15 and 20 h intervals. The samples, from different intervals, were collected in an amber bottle, purged with gas-nitrogen and stored at the temperature of approximately –18°C until the moment of analyses.

Methods

Samples submitted to thermoxidation were analyzed for fatty acid composition and total polar compounds.

Fatty acid composition

The fatty acid compositions of oil samples were analyzed using gas-liquid chromatography after transesterification. The fatty acid esters methyl (FAME) were prepared according to the procedure described by Hartman and Lago (1973). The methylation method consists of alkaline sample, followed by methylation and acid extraction with n-hexane. Weighed to 0.2 g of oil in 50 mL flask and added to 5 mL of methanol solution of potassium hydroxide 0.5 N. Then, an air condenser was connected to the balloon and the whole was heated on hotplate for 3 min. He joined the balloon, still hot, 15 mL ammonium chloride/sulfuric

Table 2 Fatty acid composition of soybean oil with LSE during thermoxidation at 180°C

	Fatty acids (%)	Heating times (h)*				
		0	5	10	15	20
	C14:0	0.08±0.01	0.23±0.05	0.39±0.02	0.35±0.01	0.39±0.02
	C16:0	10.6±0.03	10.7±0.04	10.8±0.02	10.9±0.02	11.9±0.02
* Mean ± standard deviation (n=2)	C16:1	0.14±0.03	0.22±0.04	0.23±0.02	0.31±0.02	0.31±0.02
a, b... (line)-averages followed by the same small letter do not differ by the Tukey test ($P>0.05$)	C18:0	1.9±0.02	2.9±0.02	2.9±0.03	3.1±0.02	3.2±0.01
	C18:1	20.1±0.01	21.4±0.02	22.0±0.03	23.6±0.03	24.1±0.03
	C18:2	58.3±0.02	56.9±0.01	56.3±0.01	55.0±0.03	54.1±0.03
C14:0, myristic acid; C16:0, palmitic acid; C16:1, palmitoleic acid; C18:0, stearic acid; C18:1, oleic acid; C18:2, linoleic acid; C18:3, α -linolenic acid; C20:0, arachidic acid; C20:1, eicosenoic acid; C22:0, behenic acid; AGS, saturated fatty acids; AGM, mono-unsaturated fatty acids; AGP, polyunsaturated fatty acids; NI, not identification	C18:3	6.4±0.04	5.3±0.02	5.2±0.02	4.0±0.03	3.9±0.01
	C20:0	0.03±0.04	0.09±0.02	0.16±0.02	0.20±0.01	0.22±0.02
	C20:1	0.13±0.02	0.13±0.01	0.15±0.01	0.17±0.03	0.17±0.01
	C22:0	0.41±0.01	0.48±0.03	0.49±0.01	0.49±0.01	0.50±0.03
	AGS	13.0 ^c	14.5 ^d	14.8 ^c	15.0 ^b	16.2 ^a
	AGM	20.4 ^c	21.8 ^d	22.4 ^c	24.1 ^b	24.6 ^a
	AGP	64.8 ^a	62.1 ^b	61.5 ^c	59.0 ^d	57.9 ^c
	NI	1.9	1.6	1.3	1.8	1.2

Table 3 Fatty acid composition of soybean oil with TBHQ during thermoxidation at 180°C

	Fatty acids (%)	Heating times (h)*				
		0	5	10	15	20
	C14:0	0.08±0.01	0.25±0.03	0.39±0.01	0.45±0.01	0.49±0.02
	C16:0	9.4±0.02	9.7±0.01	10.3±0.02	11.8±0.03	12.1±0.01
* Mean ± standard deviation (n=2)	C16:1	0.16±0.02	0.25±0.02	0.29±0.02	0.30±0.03	0.32±0.01
a, b... (line)-averages followed by the same small letter do not differ by the Tukey (<i>P</i> >0.05)	C18:0	1.5±0.01	2.7±0.02	2.9±0.03	3.0±0.01	3.0±0.02
	C18:1	19.4±0.03	21.5±0.02	22.3±0.04	22.8±0.02	23.3±0.02
	C18:2	59.3±0.03	58.5±0.03	56.3±0.05	54.8±0.02	54.1±0.01
C14:0, myristic acid; C16:0, palmitic acid; C16:1, palmitoleic acid; C18:0, stearic acid; C18:1, oleic acid; C18:2, linoleic acid; C18:3, α-linolenic acid; C20:0, arachidic acid; C20:1, eicosenoic acid; C22:0, behenic acid; AGS, saturated fatty acids; AGM, mono-unsaturated fatty acids; AGP, polyunsaturated fatty acids; NI, not identification	C18:3	7.9±0.02	5.5±0.01	5.2±0.04	4.6±0.01	4.1±0.03
	C20:0	0.10±0.02	0.13±0.01	0.28±0.04	0.30±0.02	0.33±0.03
	C20:1	0.08±0.02	0.10±0.03	0.13±0.03	0.14±0.01	0.14±0.01
	C22:0	0.12±0.02	0.18±0.01	0.34±0.02	0.43±0.01	0.44±0.02
	AGS	11.3 ^c	12.9 ^d	14.2 ^c	15.9 ^b	16.4 ^a
	AGM	19.7 ^c	21.8 ^d	22.8 ^c	23.2 ^b	23.8 ^a
	AGP	67.2 ^a	63.9 ^b	61.5 ^c	59.7 ^d	58.2 ^c
	NI	1.8	1.2	1.4	1.1	1.6

acid and methanol in an identical manner to methylation alkaline methylation proceeded acid. After cooling, the sample was added to 10 mL of n-hexane and stirred vigorously for about a minute. Completed the flask with a solution of chloride sodium 10%, and left to stand until complete separation of phases and phase whitening n-hexane.

The analyses of FAMES were performed with a Varian gas-liquid chromatography (Walnut Creek, USA), GC 3900 model, equipped with a flame-ionization detector, a split injector and an autosampler. FAMES were separated by using a CP-Sil 88 fused silica capillary column (50 m length, 0.25 mm internal diameter and 0.20 mm film thickness). The column oven temperature was initially held

at 50°C for 2 min, heated at 4°C/min up to 240°C and maintained at 240°C for 20.5 min. The injector and detector temperatures were 230 and 250°C, respectively. Samples of 1.0 mL were injected adopting a split ratio of 1:30. The carrier gas was hydrogen with a flow rate of 30 mL/min. FAMES were identified by comparing their retention times with those of pure FAME standards (Supelco, Bellefonte, USA) under the same operating conditions and quantified by area normalization (%).

Total polar compounds

For the determination of total polar compounds was applied the chromatographic method proposed by Dobarganes

Table 4 Fatty acid composition of soybean oil with Mixture 1 during thermoxidation at 180°C

	Fatty acids (%)	Heating times (h)*				
		0	5	10	15	20
	C14:0	0.02±0.02	0.03±0.03	0.05±0.02	0.06±0.01	0.07±0.05
	C16:0	10.5±0.02	10.5±0.03	10.6±0.02	11.5±0.04	11.9±0.06
* Mean ± standard deviation (n=2)	C16:1	0.04±0.03	0.05±0.02	0.06±0.03	0.09±0.04	0.09±0.05
a, b... (line)-averages followed by the same small letter do not differ by the Tukey test (<i>P</i> >0.05)	C18:0	2.9±0.03	2.9±0.02	3.1±0.03	3.2±0.03	3.2±0.04
	C18:1	21.2±0.03	22.2±0.02	22.2±0.03	23.5±0.03	24.2±0.03
	C18:2	57.5±0.02	57.3±0.02	57.1±0.01	55.9±0.01	54.9±0.03
C14:0, myristic acid; C16:0, palmitic acid; C16:1, palmitoleic acid; C18:0, stearic acid; C18:1, oleic acid; C18:2, linoleic acid; C18:3, α-linolenic acid; C20:0, arachidic acid; C20:1, eicosenoic acid; C22:0, behenic acid; AGS, saturated fatty acids; AGM, mono-unsaturated fatty acids; AGP, polyunsaturated fatty acids; NI, not identification	C18:3	6.4±0.02	6.3±0.02	5.9±0.01	4.6±0.04	4.2±0.04
	C20:0	0.02±0.01	0.04±0.01	0.06±0.03	0.07±0.04	0.08±0.04
	C20:1	0.10±0.01	0.12±0.02	0.13±0.02	0.17±0.02	0.17±0.01
	C22:0	0.46±0.02	0.48±0.01	0.48±0.01	0.49±0.05	0.51±0.02
	AGS	13.9 ^c	13.9 ^d	14.3 ^c	15.3 ^b	15.8 ^a
	AGM	21.4 ^c	22.1 ^d	22.5 ^c	23.8 ^b	24.5 ^a
	AGP	63.9 ^a	63.6 ^b	63.0 ^c	60.5 ^d	59.0 ^c
	NI	0.82	0.29	0.28	0.45	0.65

Table 5 Fatty acid composition of soybean oil with Mixture 2 during thermoxidation at 180°C

	Fatty acids (%)	Heating times (h)*				
		0	5	10	15	20
* Mean $\pm$ standard deviation ($n=2$)	C14:0	0.01 $\pm$ 0.02	0.02 $\pm$ 0.01	0.02 $\pm$ 0.04	0.03 $\pm$ 0.01	0.03 $\pm$ 0.03
a, b... (line)-averages followed by the same small letter do not differ by the Tukey test ($P>0.05$)	C16:0	10.6 $\pm$ 0.01	10.6 $\pm$ 0.06	10.7 $\pm$ 0.04	11.9 $\pm$ 0.01	12.3 $\pm$ 0.03
	C16:1	0.02 $\pm$ 0.03	0.05 $\pm$ 0.03	0.06 $\pm$ 0.03	0.06 $\pm$ 0.01	0.06 $\pm$ 0.01
	C18:0	2.3 $\pm$ 0.03	2.9 $\pm$ 0.06	3.1 $\pm$ 0.03	3.1 $\pm$ 0.01	3.1 $\pm$ 0.01
	C18:1	21.3 $\pm$ 0.04	22.2 $\pm$ 0.02	22.5 $\pm$ 0.02	22.9 $\pm$ 0.01	23.3 $\pm$ 0.02
	C18:2	57.7 $\pm$ 0.02	56.8 $\pm$ 0.01	56.5 $\pm$ 0.02	55.9 $\pm$ 0.01	55.8 $\pm$ 0.04
	C18:3	6.4 $\pm$ 0.03	6.3 $\pm$ 0.03	5.8 $\pm$ 0.01	4.7 $\pm$ 0.01	4.5 $\pm$ 0.04
	C20:0	0.02 $\pm$ 0.05	0.02 $\pm$ 0.03	0.03 $\pm$ 0.02	0.04 $\pm$ 0.01	0.04 $\pm$ 0.05
	C20:1	0.08 $\pm$ 0.05	0.10 $\pm$ 0.02	0.10 $\pm$ 0.02	0.12 $\pm$ 0.01	0.13 $\pm$ 0.07
	C22:0	0.46 $\pm$ 0.01	0.48 $\pm$ 0.02	0.48 $\pm$ 0.03	0.49 $\pm$ 0.01	0.50 $\pm$ 0.05
	AGS	14.0 ^c	14.1 ^d	14.3 ^c	15.6 ^b	16.0 ^a
	AGM	21.4 ^c	22.4 ^d	22.6 ^c	23.1 ^b	23.5 ^a
	AGP	64.1 ^a	63.1 ^b	62.3 ^c	60.7 ^d	60.3 ^c
	NI	0.40	0.46	0.77	0.70	0.20

et al. (2000), using hexane: ethyl ether 90:10 to produce better separation of the non-polar. The content of polar compounds, polar fraction was calculated from triacylglycerols were not altered, whereas retained polar compounds were included in the polar fraction. The results obtained by column chromatography, were expressed in percentages.

Statistical analysis

The test was performed in entirely casual delineation (Gacula and Singh 1984). Variance analysis and Tukey test at 5% were obtained through the ESTAT-Sistema para Análises Estatísticas (Statistical Analyses System), software version 2.0.

Results and discussion

Fatty acid composition

The mean results of percentage composition in fatty acids for treatments can be observed in Tables 1, 2, 3, 4 and 5. Note that the initial samples used in this study presented in fatty acid composition in accordance with Brazilian law states for refined vegetable oils (Codex Alimentarius Commission 2008).

When evaluating fatty acids profile for treatments during the heating process, there was a significant difference ($P<0.05$) with increased percentage of saturated and mono-unsaturated fatty acids and decreased the amount of polyunsaturated, considered essential fatty acids.

Table 6 Total polar compounds (%) for the different treatments over the heating process to 180°C

Treatments	Heating times (h)*				
	0	5	10	15	20
Control	4.43 $\pm$ 0.12 ^{eA}	7.43 $\pm$ 0.16 ^{dA}	17.94 $\pm$ 0.21 ^{4cA}	35.50 $\pm$ 0.27 ^{bA}	40.37 $\pm$ 0.32 ^{aA}
LSE	2.72 $\pm$ 0.31 ^{eB}	5.83 $\pm$ 0.14 ^{dB}	15.61 $\pm$ 0.18 ^{cB}	25.81 $\pm$ 0.24 ^{bB}	35.51 $\pm$ 0.28 ^{aB}
TBHQ	3.41 $\pm$ 0.22 ^{eAB}	5.67 $\pm$ 0.23 ^{dB}	16.63 $\pm$ 0.35 ^{cB}	34.93 $\pm$ 0.32 ^{bA}	40.98 $\pm$ 0.40 ^{aA}
Mixture 1	3.61 $\pm$ 0.25 ^{cAB}	3.77 $\pm$ 0.18 ^{cC}	11.85 $\pm$ 0.32 ^{bC}	22.11 $\pm$ 0.48 ^{aC}	22.43 $\pm$ 0.20 ^{aC}
Mixture 2	3.72 $\pm$ 0.15 ^{cAB}	3.85 $\pm$ 0.31 ^{cC}	11.99 $\pm$ 0.43 ^{bC}	22.24 $\pm$ 0.33 ^{aC}	22.32 $\pm$ 0.22 ^{aC}

* Mean $\pm$ standard deviation ($n=2$)

Control: soybean oil; LSE: lemon seeds extract (2,400 mg/kg); TBHQ: tertiary-butyl hydroquinone (50 mg/kg); Mixture 1: LSE (2,400 mg/kg) + TBHQ (50 mg/kg); Mixture 2: LSE (2,400 mg/kg) + TBHQ (25 mg/kg)

a, b... (line)-averages followed by the same small letter do not differ by the Tukey test ($P>0.05$)

A, B... (column)-averages followed by the same capital letter do not differ by the Tukey test ($P>0.05$)

This way, we could observe that at the end the heating process the percentages of saturated fatty acids were increased by 52.31, 45.43, 24.56, 14.17 and 13.75% for the control, ESL, TBHQ, Mixtures 1 and 2, respectively. The same trend was observed for mono-unsaturated fatty acids, resulting an increase of 31.11% for control, 20.62% for ESL, 21.02% for TBHQ, 14.45 and 9.52% for Mixture 1 and 2, respectively. As for the essential fatty acids, there was a decrease in linoleic and linolenic acids in a higher percentage to the control (17.47%), followed by LSE (10.46%), TBHQ (13.43%), Mixture 1 (7.56%) and Mixture 2 (5.97%).

According Pantzaris (1998), the decrease in content of linoleic and linolenic acids during the heating process is due to their destruction by oxidation, polymerization, among others and should, therefore, be an important quality test for the oils.

Total polar compounds

Table 6 shows the total polar compounds means for different treatments throughout the heating process. From 5 h until the end of heating, it is observed that the treatments added to antioxidant substances showed protective action of oil on the formation of the total polar compounds, although at different levels of efficiency.

It appears that at 5 h, the oil added to Mixtures 1 and 2 showed lower formations of polar compounds. At 20 h of heating the treatments LSE, Mixtures 1 and 2 delayed the formation of polar compounds to 12.03, 44.44 and 44.71% respectively, showing efficiency in protecting the oil. It is noteworthy that the TBHQ showed similar behavior to Control, not delaying the formation of total polar compounds.

In Brazil there is no law establishing the maximum amount of polar compounds in frying oils. After 20 h at 180°C, the oil added to LSE over the limit of 25% (35.51%), was recommended by international law. However, this has not occurred with treatments that were added to Mixtures 1 and 2, which reached approximately 22% of the total polar compounds. The TBHQ, in the case of a synthetic antioxidant widely used in oil under high temperatures, exceeded this limit, showing about 41% of polar compounds, when added at a concentration of 50 mg/kg.

In general, the values obtained for polar compounds in this study, are close to those cited in the literature. Thus, soybean oil showed 17.94% of polar compounds with 10 h of thermoxidation at 180°C, while Barrera-Arellano et al. (2002) obtained 18.6%.

Results demonstrate that, despite of the treatments studied, there was an increased percentage of saturated and mono-unsaturated fatty acids, and reduction of poly-unsaturated fatty acids. As for total polar compounds, findings show that the LSE presented efficiency in

protecting the oil when added alone to soybean oil submitted to thermoxidation. However, treatments Mixtures 1 and 2 had the highest antioxidant power applied alone, thus proving the synergistic effect of the antioxidants studied.

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